

Serial No.: 09/763,957
Docket No.: 229752001300
Inventor: Jose Ramon Botella MESA et al.
Title: A PLANT PROMOTER AND USES THEREFOR
(as amended)
Substitute Sheet 1 of 28

AIM-1 OLIGONUCLEOTIDES

-Oligonucleotide primers (SEQ ID NOS:4-9) used during Long Distance Inverse PCR
-Oligo's bind to regions of AIM-1 (Mung bean ACC Synthase)

NSE-1 to NSE-6 correspond to SEQ ID NOS:4-9, respectively.

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NSE-1

5' -GC GGAT↓CCATCTTGGACAACAAGGGAGTT - 3'

29'omer

Tm = 68

NSE-2

5' -TAGGATC↓CAGAAAGACACTGAGAACCGTGG- 3'

30'omer

Tm = 70

NSE-3

5' -ACGGATCC↓GGTGTATGTGGTTAGAGTGTG- 3'

29'omer

Tm = 62

NSE-4

5' -CAGGATC↓CAGACATAGAGTGTGACCGCAA- 3'

29'omer

Tm = 66

NSE-5

5' -ATCGATCATATGAGCTCTAGACCCGGCTGCAGGATCC↓GGTGTATGTGGTTAGAGTGTG- 3'

59'omer

Tm = 62

note: NSE-5 is identical to NSE-3 except different restriction enzyme sites have been incorporated
(ie. 5'-Cla I, Nde I, Sac I, Xba I, Sma I, Pst I & Barn HI-3')

NSE-6

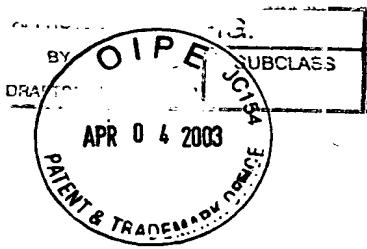
5' -CCGCGGAGATCTATCGATCTCGAGAATTCAAGCTT↓CAGACATAGAGTGTGACCGCAA-3'

57'omer

Tm = 66

note: NSE-6 is identical to NSE-4 except different restriction enzyme sites have been incorporated
(ie. 5'-Sac II, Bgi II, Cia I, Xho I, Eco RI, & Hind III-3')

FIGURE 1



Serial No.: 02/063,957
Docket No.: 152001300
Inventor: Jose Ramon Botella MESA et al.
Title: A PLANT PROMOTER AND USES THEREFOR
(as amended)
Substitute Sheet 2 of 28

2/28

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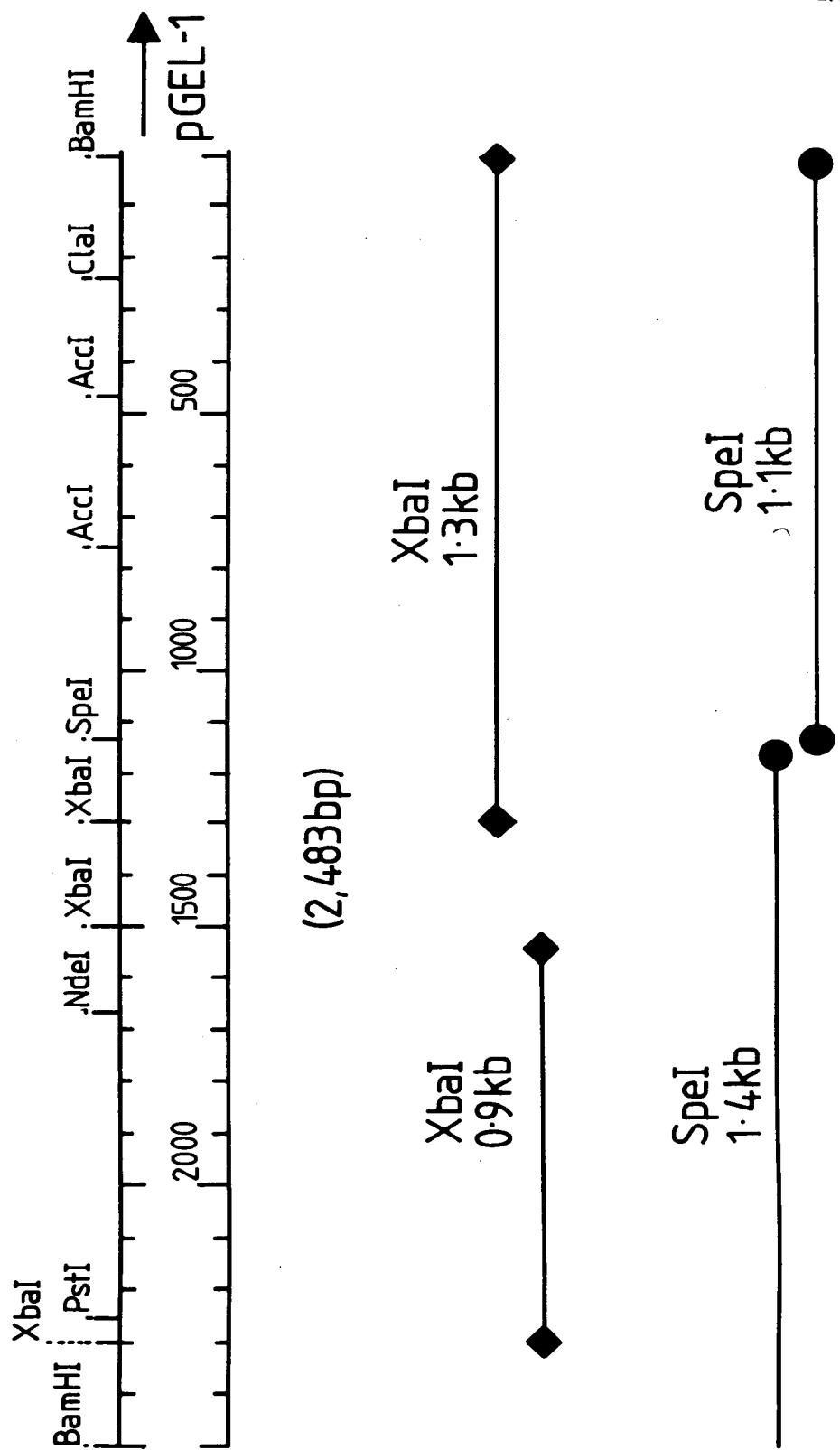


FIGURE 2

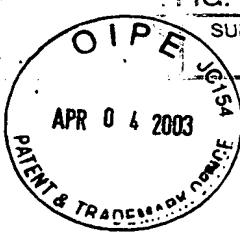


FIG.

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Serial No.: 09/763,957
 Doc. No.: 229752001300
 Inventor: Rose Ramon Botella MESA et al.
 Title: A PLANT PROMOTER AND USES THEREFOR
 (as amended)
 Substitute Sheet 3 of 28

3/28

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pGEL-1 2.5Kb promoter sequencing strategy:

A11 fragments

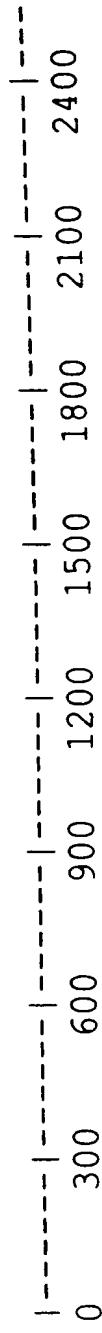
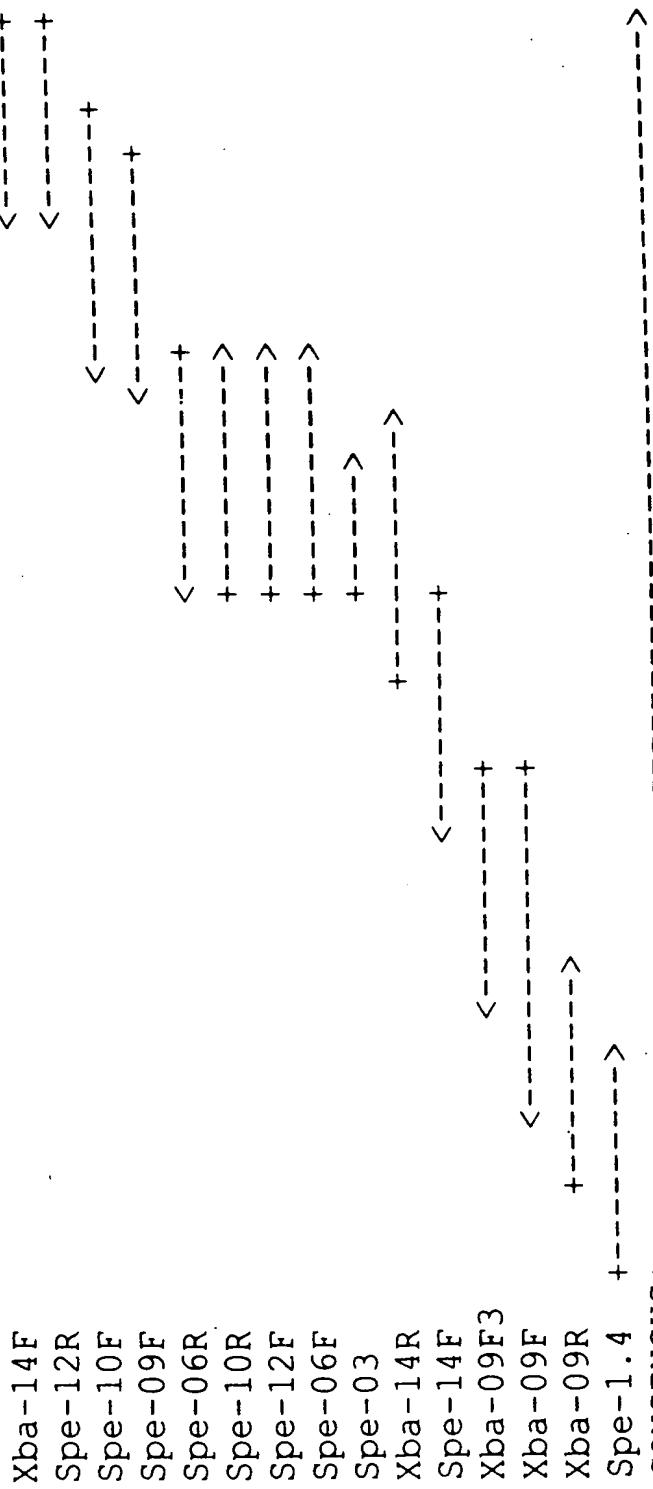
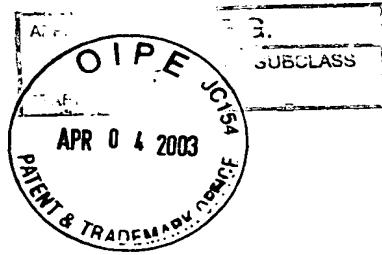


FIGURE 3



Serial No.: 09/763,957
Docket No.: 229752001300
Inventor: Rose Ramon Botella MESA et al.
Title: A PLANT PROMOTER AND USES THEREFOR
(as amended)
Substitute Sheet 4 of 28

4/28

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APR 09 2003

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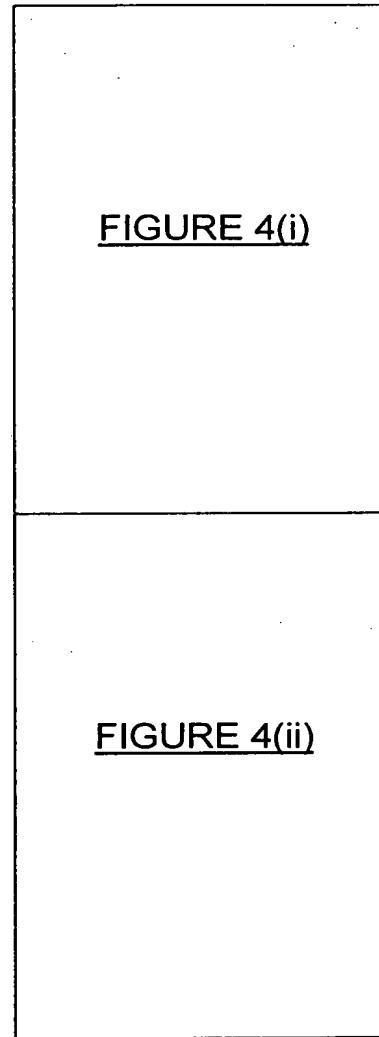


Figure 4 corresponds to SEQ ID NO:3

FIGURE 4



5/28

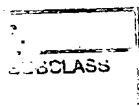
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pGEL-1 2.5Kb Promoter Fragment

Length: 2470

1 TTACAGATAC ACAGAACATC ACGACACATC TACTTTAATA ACAGAAAAAT
51 AATAAGTGTC GGAGATTATG GTACGACAAG ATGAAATGTT TTTATATGGT
101 TGAGATTATT TTGGTCTGTT GTTGGAAAGTT TCACGAATCA TGATTTGAT
151 TTTACGTATT AAAAAATGAA AAGTTGAATC ATGCATTTA TCTAGAAGCT
201 GGGAACTGAA CCAAAAAAAT AGCCAGTTGA ACAACTGCAG TATTTGTAGG
251 CGTATTCAATT TCTCCTTCC TACAATAATC CTTGGTTGCT CTTTATCGGA
301 AAAAAACCAA AAGCAATAGC TACTCTGTAA GGTCCTCGAT TGCCGACAAG
351 AACATCACAT GCGTGCTGTC GAAGAACACA TAATTTGAG GTTGAAGCTC
401 ACGTGCAGT TTTGCATATT TTTAGGTTAT GTGTACACGT ATGGAGTGAG
451 TTCCCGGTAT ATAGTGTAGG TAGTTGAGTG GCTGAGTAGC GAGTGAATCA
501 GGTAAACACTA TCTTTCAAG CCACCTAATT AAGGGATTAA ATGTTCATGC
551 AACTGTTCTT CGCTAACTAA GGCCCCACTT ACCTTTATAA TATTCTCTCT
601 AACTCCGGGC TTTGGTAAG TACAACCTTT CTACTCTTAT TTAATGGAGG
651 GATTATTTT TCCATATAACC AATTAATTAA TTTTTAATT TATGCATTTT
701 GATCTTATAT TAAAACAATT ATGGTATGGA TTAAGTCGTA TATCGGTGAC
751 AATTGAAGTT TTCCCTCAAGT TTAGGCCATT TTATGAAATT AAACCTTAATC
801 ACTACTATTA GGTAAATTCA TATGTATCAT TAACAATTTC AATGTGAGTT
851 CAATTTACC CAAGATTGA AAGTTGTTGT CAACTTCTGT TAACTAAAGT
901 TGTATTATAA GGTTGACGAC TTTAACCTAA ATCTATTTG AATTGAAGGG
951 GTTGATGACT TCAGCTTAA AATAATTCAA CTAAAGTTCT AGACTACATT
1001 GGAGATTAA GTGTTCAAA AATTTAGAA AAAGGCTGAG TTAAAGTTAT
1051 GAAAAAGATT GGTGACTATT CAATTAATTAA GTTGTGAATT GATGACAAAT
1101 ATTTCATGAG CATAACCAAT CAGAGAAATA CCACCTCGAC CGACTACAAC
1151 AATCTCAATG TTAATTAATG AAGCATTGTA GTATAAGGAG TCTAGAATAA
1201 ATTTCTTAAA TATTAGAGGA AAACATTTT TAAAAAATTAA CAAGAAAAGT
1251 TTGATCTATA ACCTCTTAA ACTTTAAATT ATCTAACAAAT TTTCTTATGA

FIGURE 4(i)



Serial No.: 09/763,957
Docket No.: 229752001300
Inventor: Rose Ramon Botella MESA et al.
Title: A PLANT PROMOTER AND USES THEREFOR
(as amended)
Substitute Sheet 6 of 28

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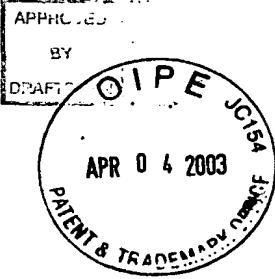
APR 09 2003

TECH CENTER 1600/2000

6/28

1301 CTCACATTGT GTTGATAGGG TGATTTGTC AAAATATATG TCTATTTAT
1351 ACTAGTATGA TTTGTCTGCG AATTATATAT AGTATTAAC TGGAGAAATG
1401 ATTGCCTAAT AAGTTATAAA AAAGGAGAAA ATATTTATTC ATAAAAAAA
1451 TACACTTAAA TAAGTAACAA TAATAAAAAA CATTATATAA GAGATTAAGA
1501 TAATTTAATA AGTATTGAAT GTAGAATAAT TTTTATTTAT AAATTGAAC
1551 TAAAATATTC AAATAATATT CAAAGTAAAT AATAGATATA ATTCACTCATT
1601 CAATACGAGT AATTCAATCT ATTATAATCC ATATATTAGA TAAATATACA
1651 AATATTTGTT AAATTTACA TTATTATATT ACTAAATATA TATTAATTTC
1701 CTTTGAATAT CTTTATACA AGTAGGTAGA CTAGAAGAAT TATCTTATCT
1751 CCCGTATATT TGTAGATGTT AAATGTAACG GGCTTAGACT GATGTTTTG
1801 TATTATATTA TTTATAAATC CATTAGAGAT TTAAGTTAAT GTCTCTCTT
1851 GATTTAAAC ATGGTCTAAA AATTAGGTTT AATCATTGCG TCCTCAATGA
1901 ACCCATGCTA TATGTTTAA AGTTTTGT TTTTGACAA TGTTTTTAT
1951 TTCTGAGATT GCTCTTAGGA TTGAAATTAT GTTGATACT AGAAAACGAA
2001 GAAGTAGAGA GTAGTGTATA CACGTGTAAA AAATAATAGT TGTGGAACT
2051 TAAGTTGGAT TTGAATACTA GGACGGAGGCT GGAAGGGTTT CCACTAAGTT
2101 GACAAAAATT ATTACAAGTG GCAACTAGCT AGGTCTCACA AAGTATTACT
2151 AATTAATAGT GGGTCTGTCT GCATACCAAC TCTTGCCTAA TTTCAAACA
2201 CCGCATTCTC TCTTCTTCTC TCCTCTTCC TCTGGAAACT TCATCGATGT
2251 GGACTTCTGT CTCTAAAAG TCAAGCTCAA TTTATCCAAT GCATTATAAA
2301 TACACACTCT CCCTCCCTTC TATTCTTCAT TGCATCACAT TTCCCTCTATA
2351 AATTACTCAC ACCTTATTCC TAACTTCATT TCAACATCCT CTCTCCACT
2401 TACTTCGATT TCATCAATTG CAATAAACTC AACACACTTT TTTACACTCC
2451 ACACCTAAAC CACATACACC

FIGURE 4(ii)



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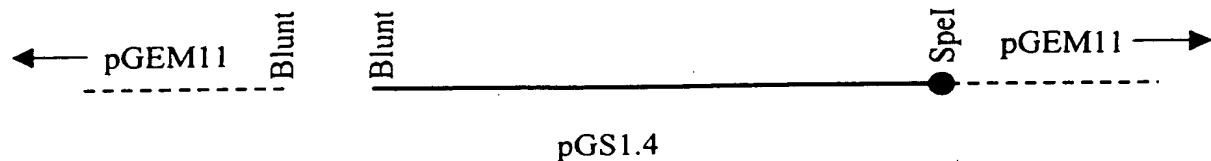
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7/28

Reconstruction of 2.5 kb pGEL-1 promoter

(a)

1. Cut HindIII and blunt end pGS1.4

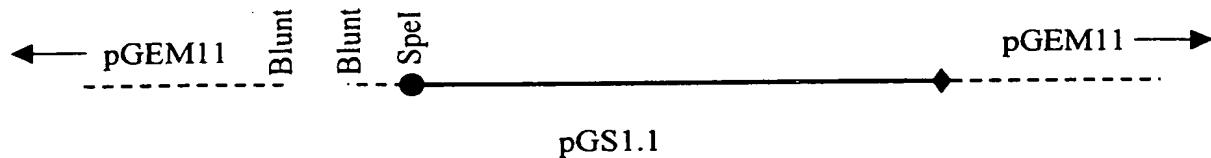


2. Cut SpeI

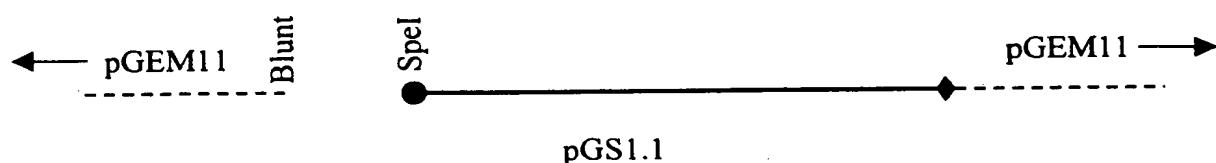


(b)

1. Cut SalI and blunt end pGS1.1



2. Digest with SpeI



(c) Ligate (a) into (b)

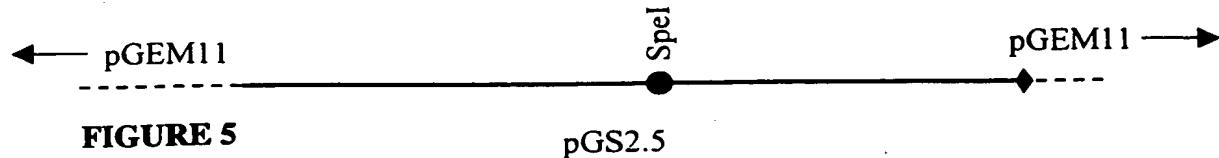


FIGURE 5

pGS2.5

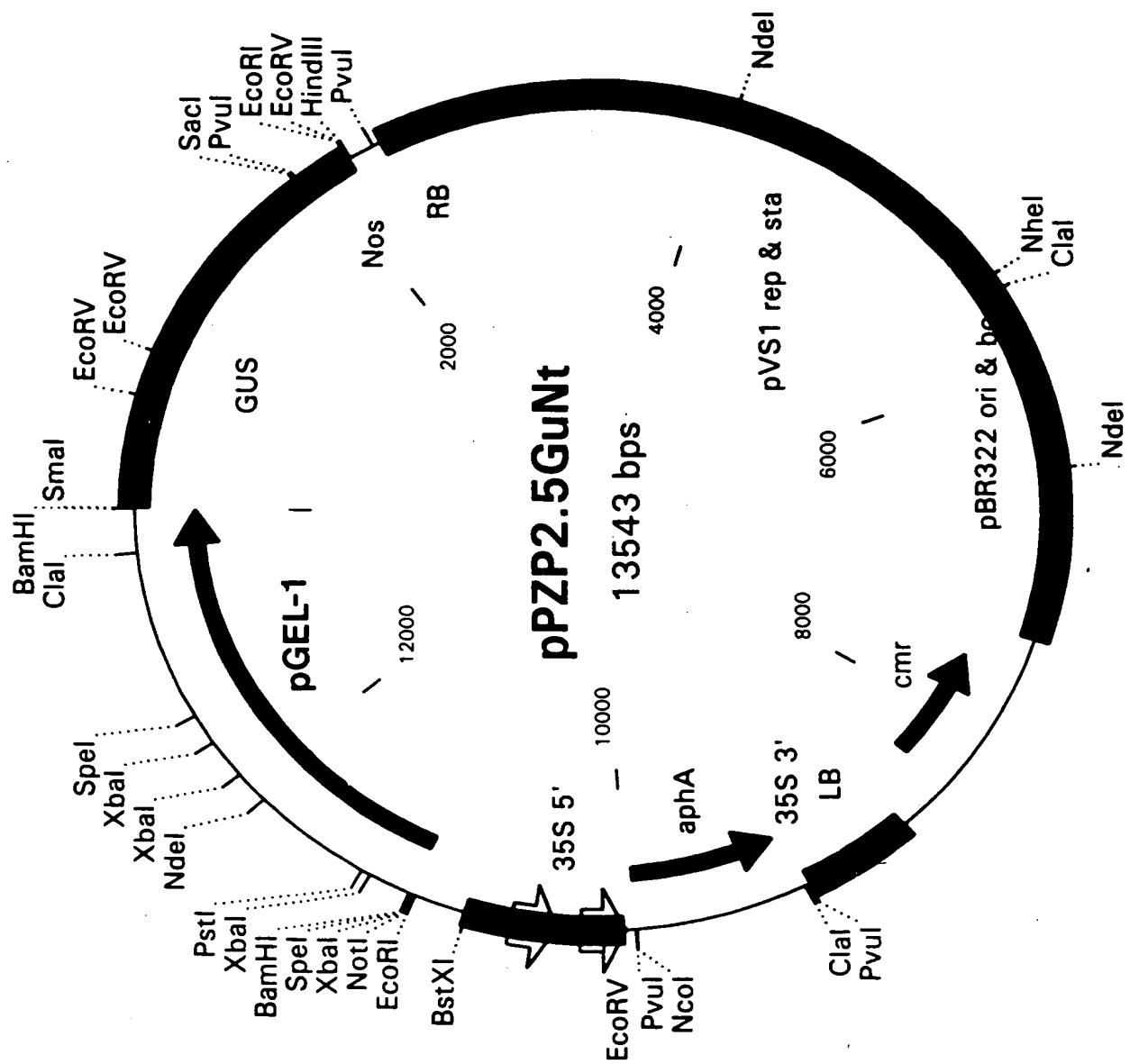
8/28

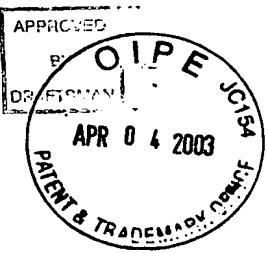
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FIGURE 6A(i)

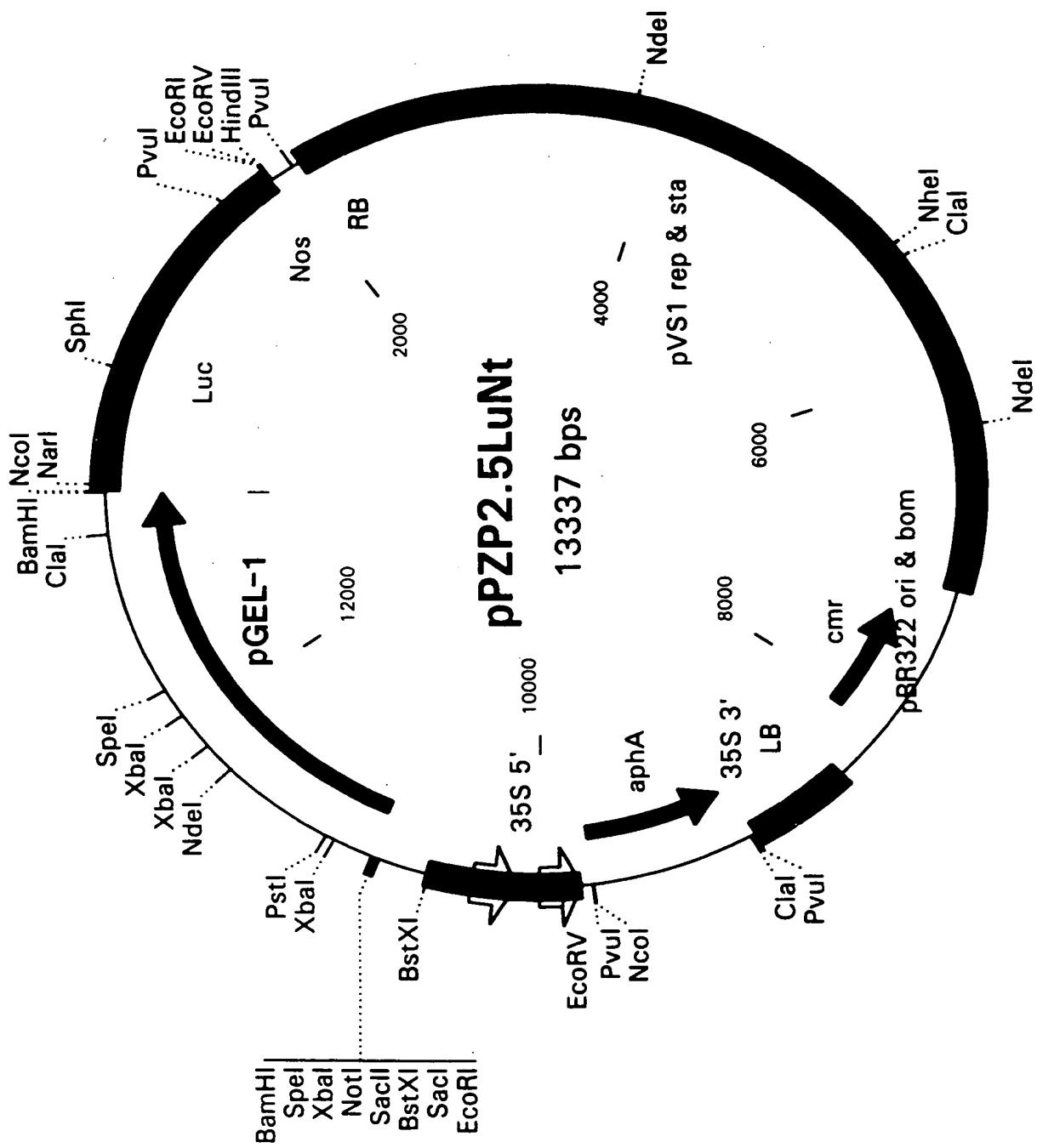




Serial No.: 09/763,957
Docket No.: 229752001300
Inventor: Rose Ramon Botella MESA et al.
Title: A PLANT PROMOTER AND USES THEREFOR
(as amended)
Substitute Sheet 9 of 28

9 / 28

FIGURE 6A(ii)



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APR 09 2003

TECH CENTER 1600/2000

10 / 28

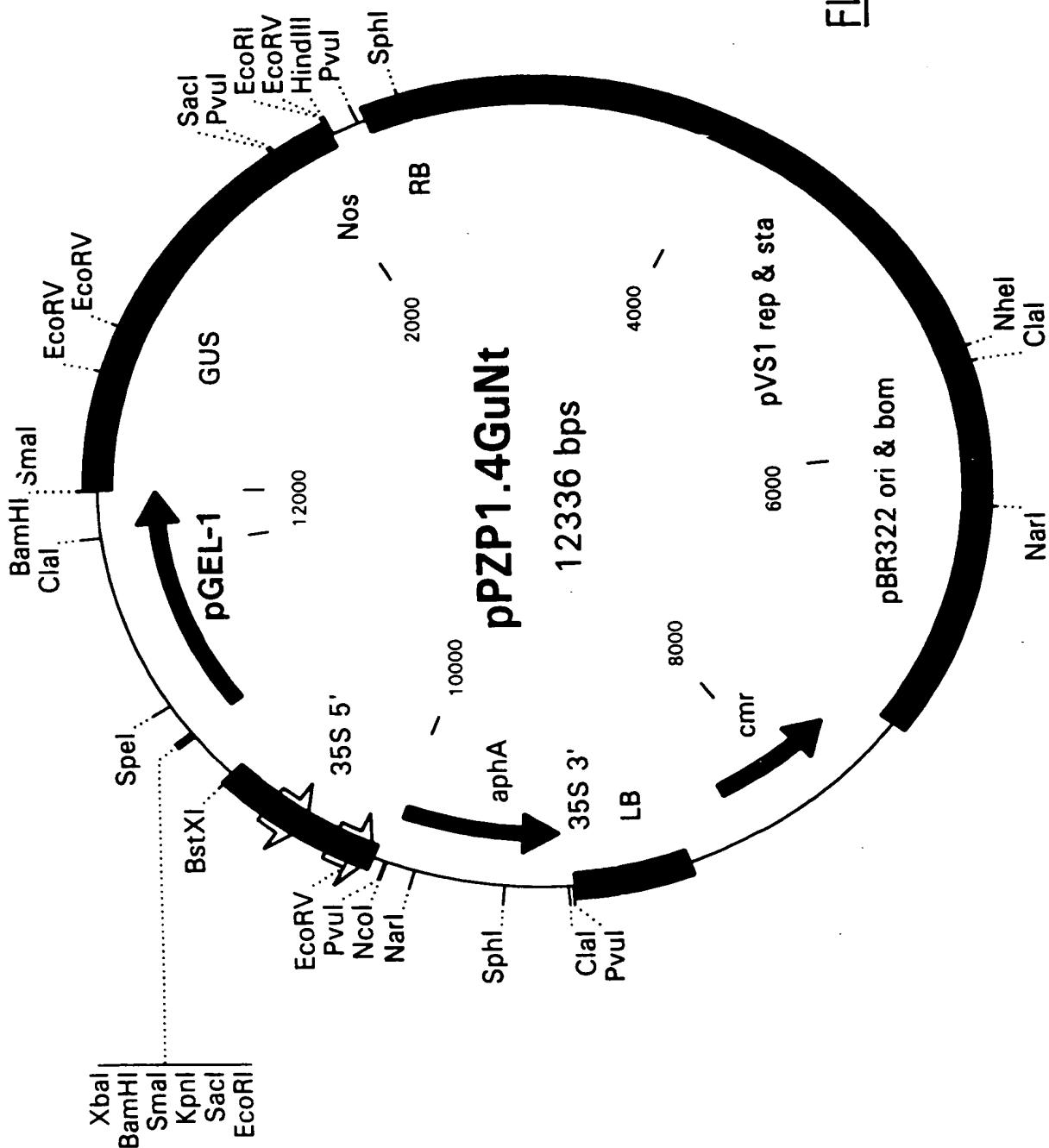


FIGURE 6A(iii)



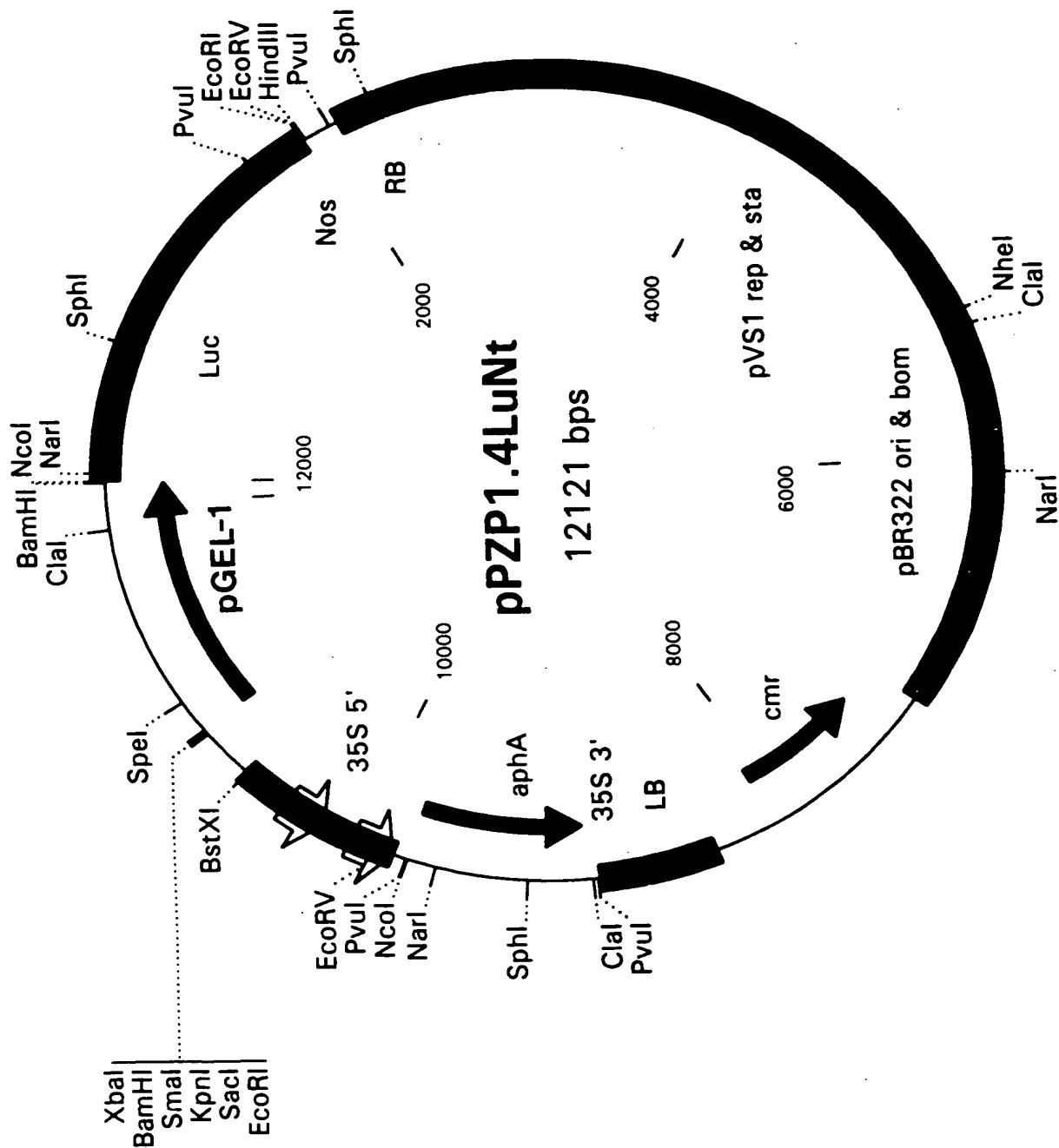
11/28

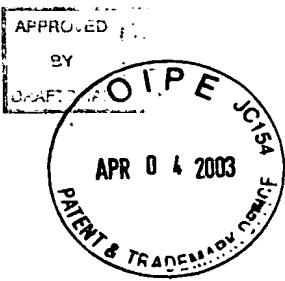
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APR 09 2003

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FIGURE 6A (iv)





Serial No.: 09/763,957
Docket No.: 229752001300
Inventor: Jose Ramon Botella MESA et al.
Title: A PLANT PROMOTER AND USES THEREFOR
(as amended)
Substitute Sheet 12 of 28

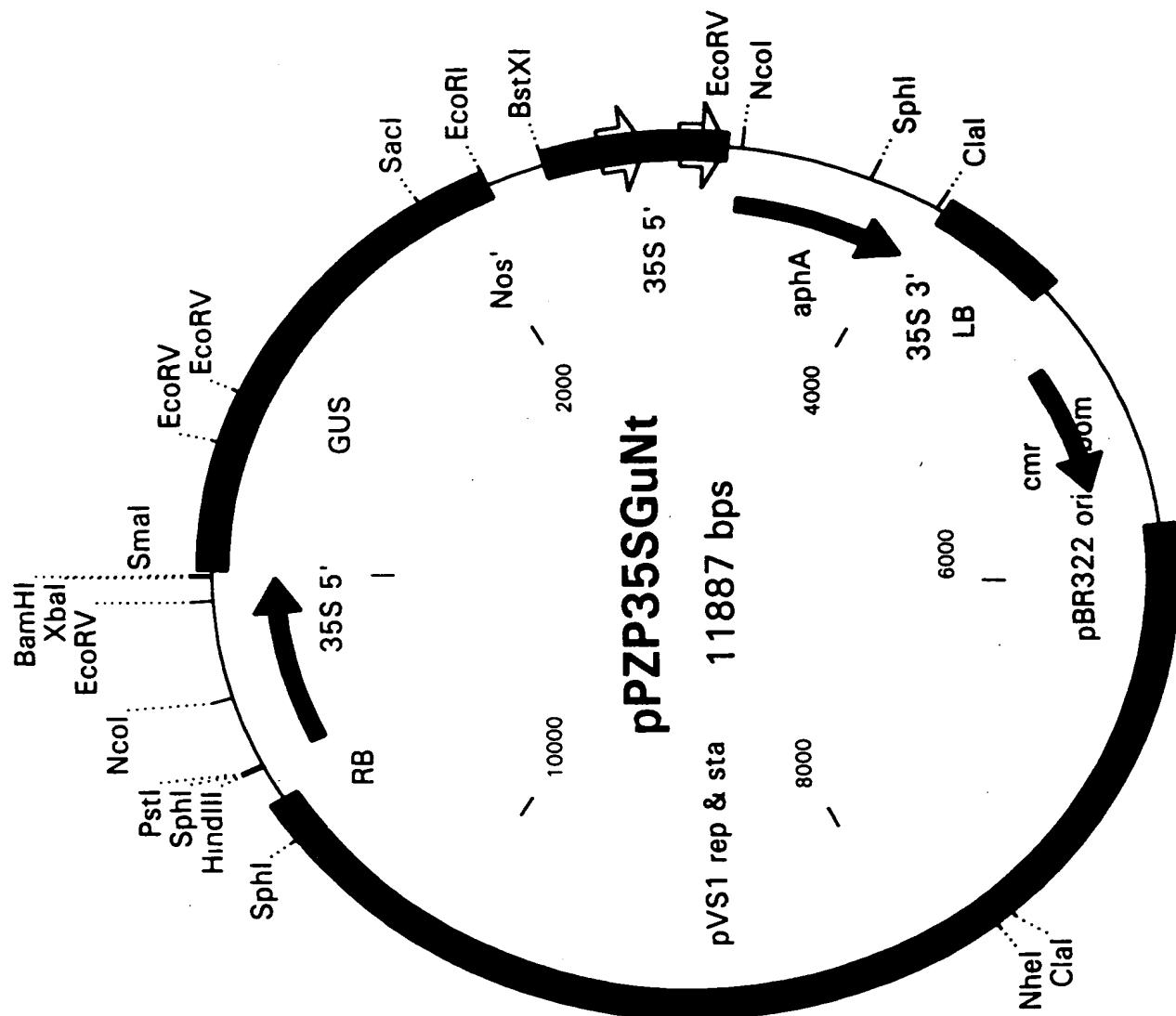
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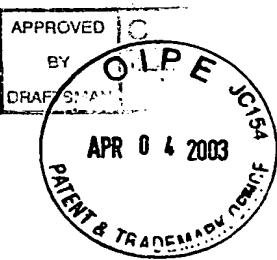
12 / 28

APR 09 2003

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FIGURE 6A(v)





Serial No. 07763,957
Docket No. 9752001300
Inventor: Jose Ramon Botella MESA, et al.
Title: A PLANT PROMOTER AND USES THEREFOR
(as amended)
Substitute Sheet 13 of 28

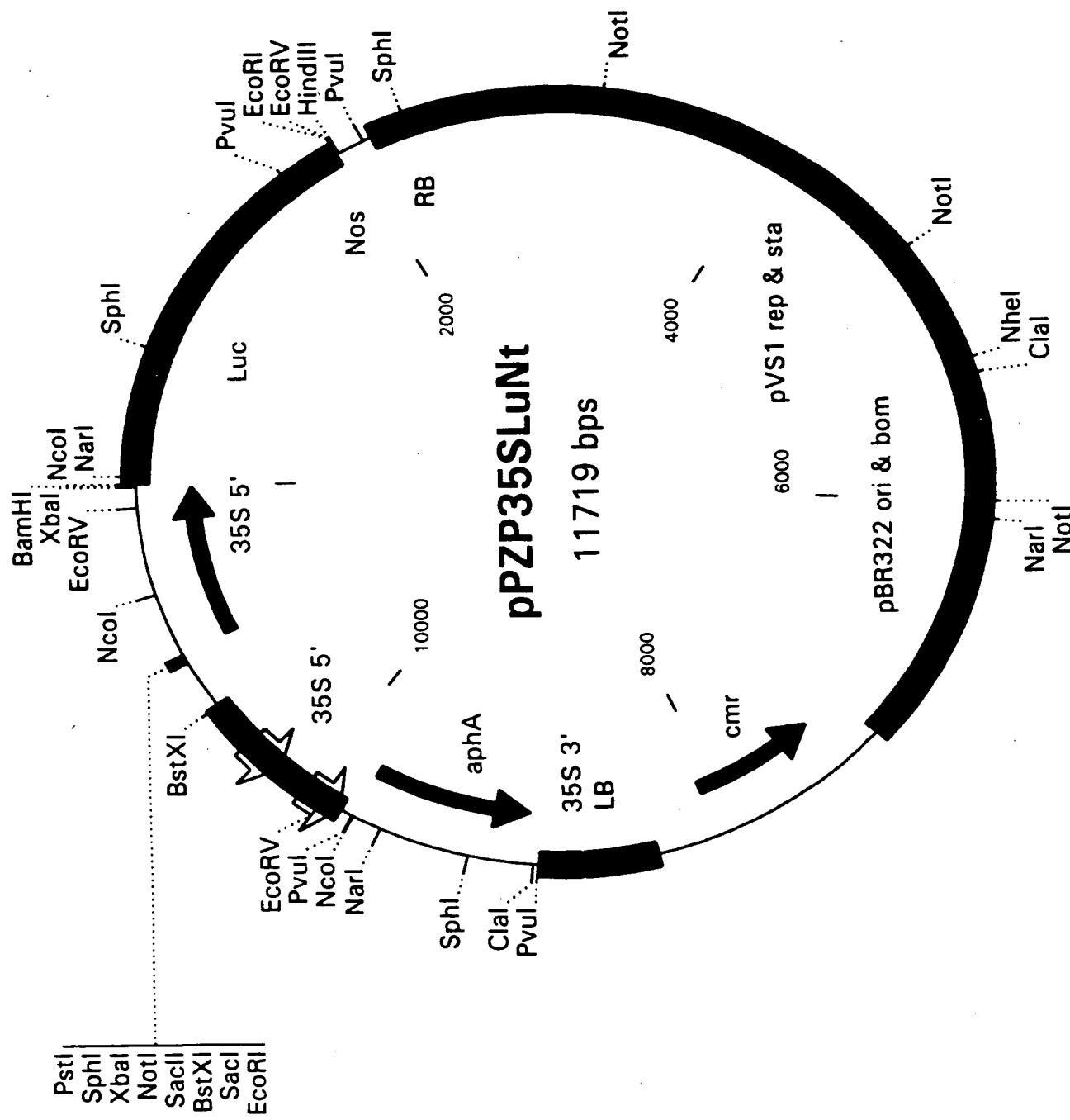
13 / 28

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APR 09 2003

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FIGURE 6A(vi)





Serial 09/763,957
 Dock 229752001300
 Inventor: Jose Ramon Botella MESA et al.
 Title: A PLANT PROMOTER AND USES THEREFOR
 (as amended)
 Substitute Sheet 14 of 28

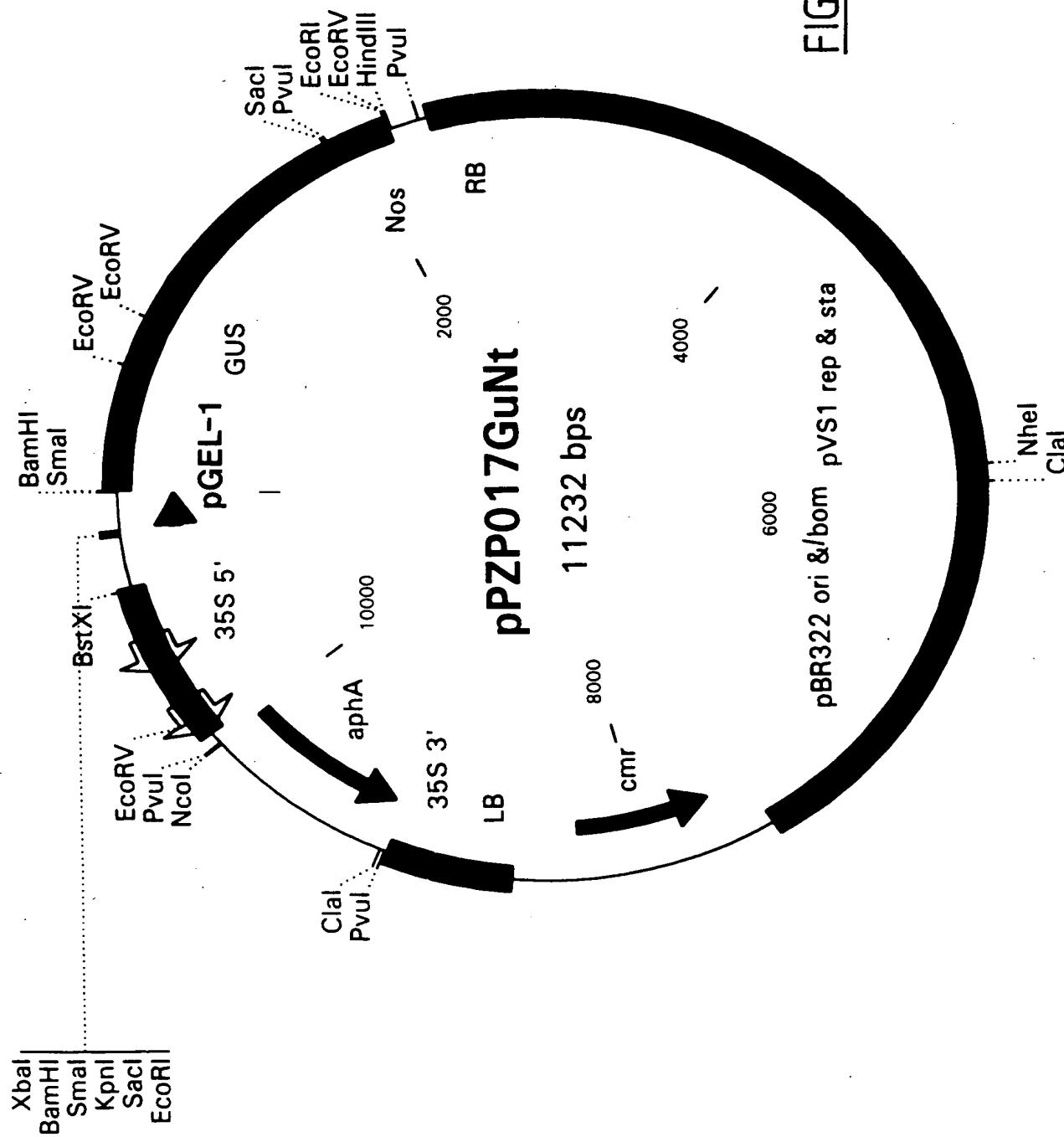
14/28

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FIGURE 6A(vii)



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APR 09 2003

TECH CENTER 1600/2800

15 / 28

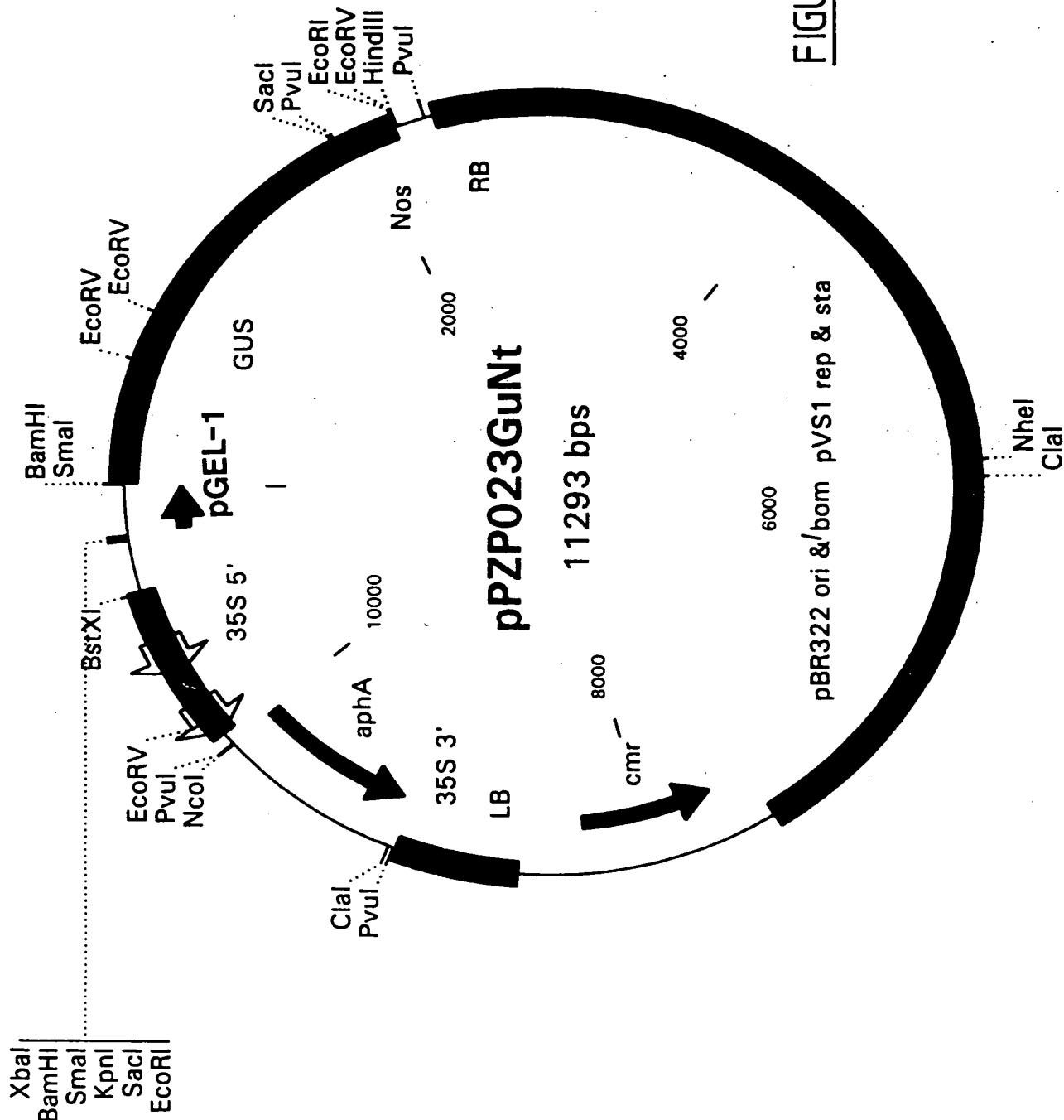


FIGURE 6A(viii)

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APR 09 2003

TECH CENTER 1600/2000

16/28

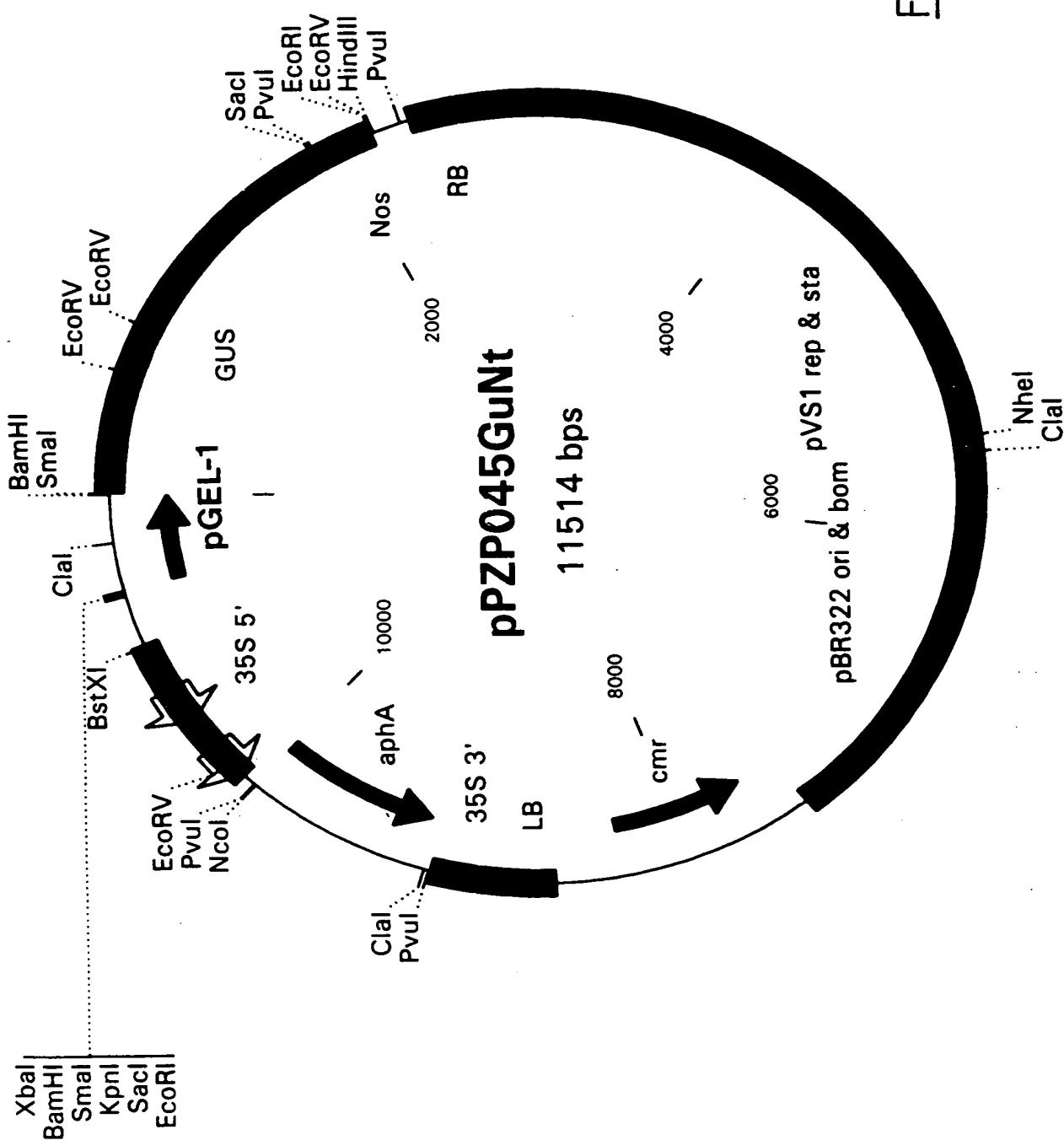
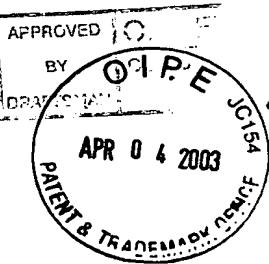


FIGURE 6A(ix)



Serial No. 09/763,957
 Docket No. 229752001300
 Inventor: Rose Ramon Botella MESA et al.
 Title: A PLANT PROMOTER AND USES THEREFOR
 (as amended)
 Substitute Sheet 17 of 28

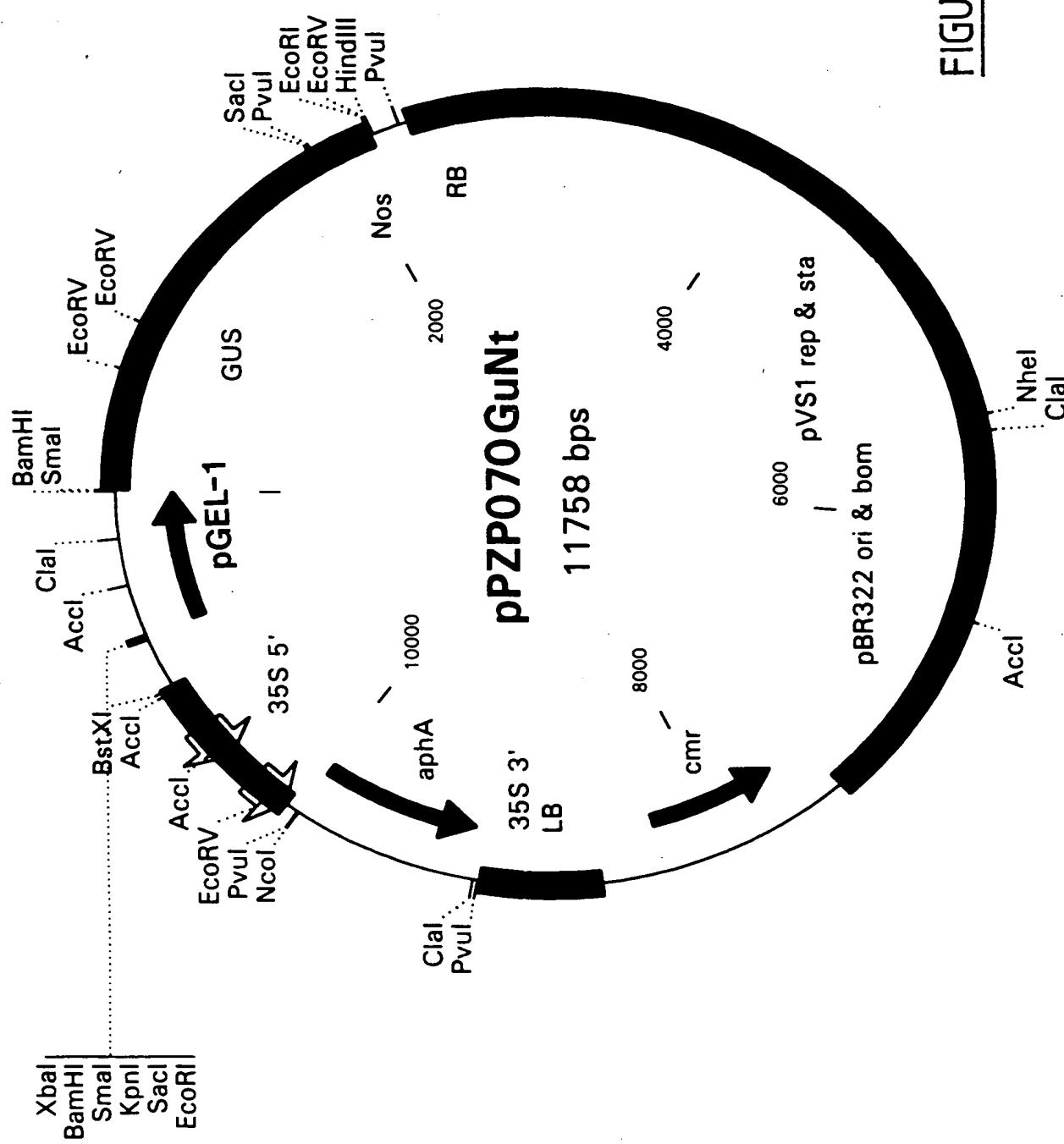
17 / 28

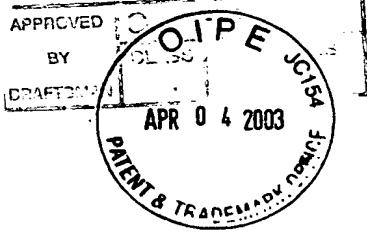
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APR 09 2003

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FIGURE 6A(X)





Serial No.: 09/763,957
Filing No.: 229752001300
Inventor: Rose Ramon Botella MESA et al.
Title: A PLANT PROMOTER AND USES THEREFOR
(as amended)
Substitute Sheet 18 of 28

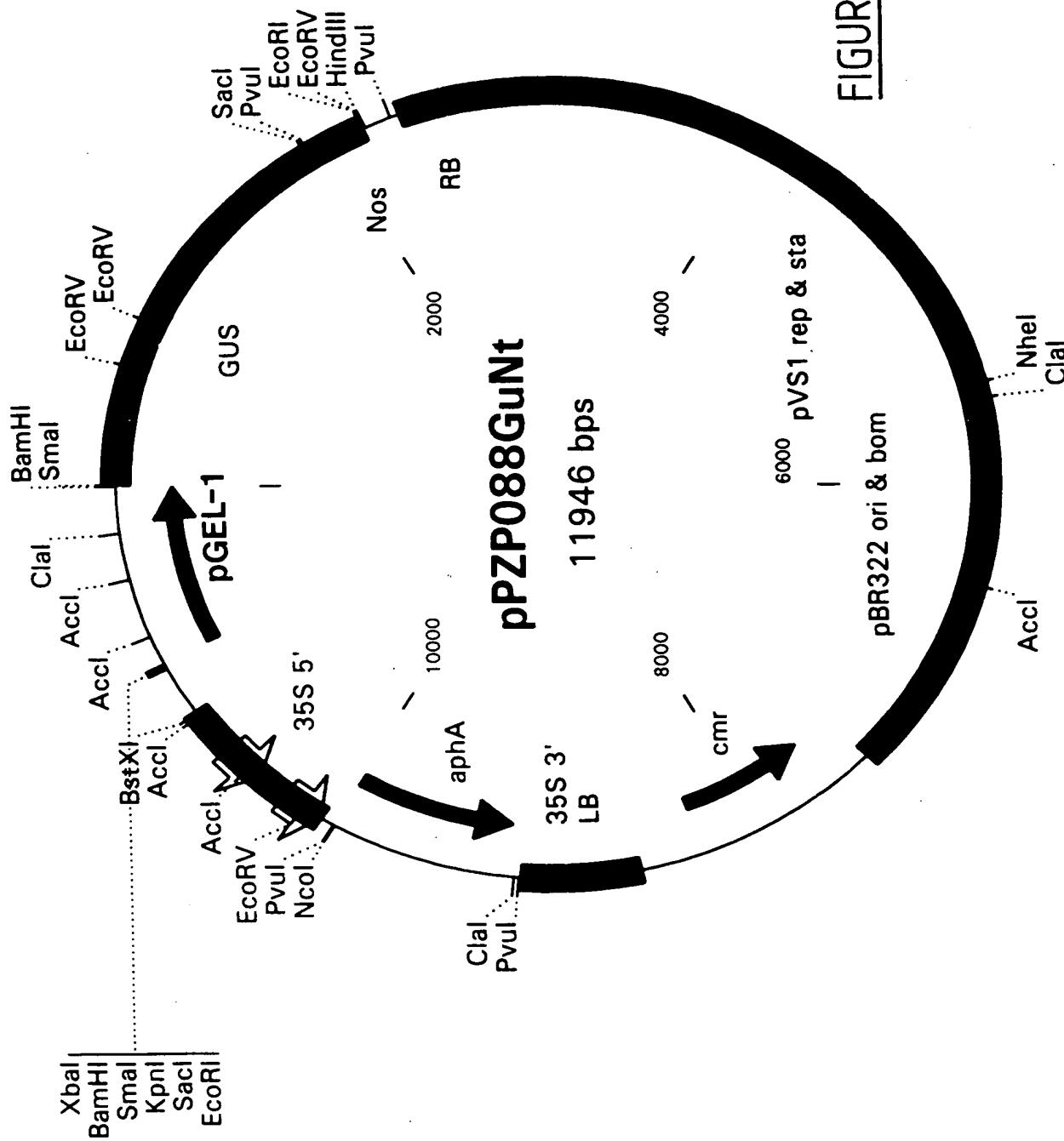
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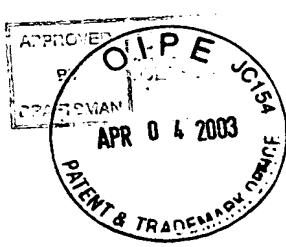
APR 09 2003

TECH CENTER 1600/2000

18 / 28

FIGURE 6A(xi)





Serial No.: 09/763,957
Docket No.: 229752001300
Inventor: Rose Ramon Botella MESA et al.
Title: A PLANT PROMOTER AND USES THEREFOR
(as amended)
Substitute Sheet 19 of 28

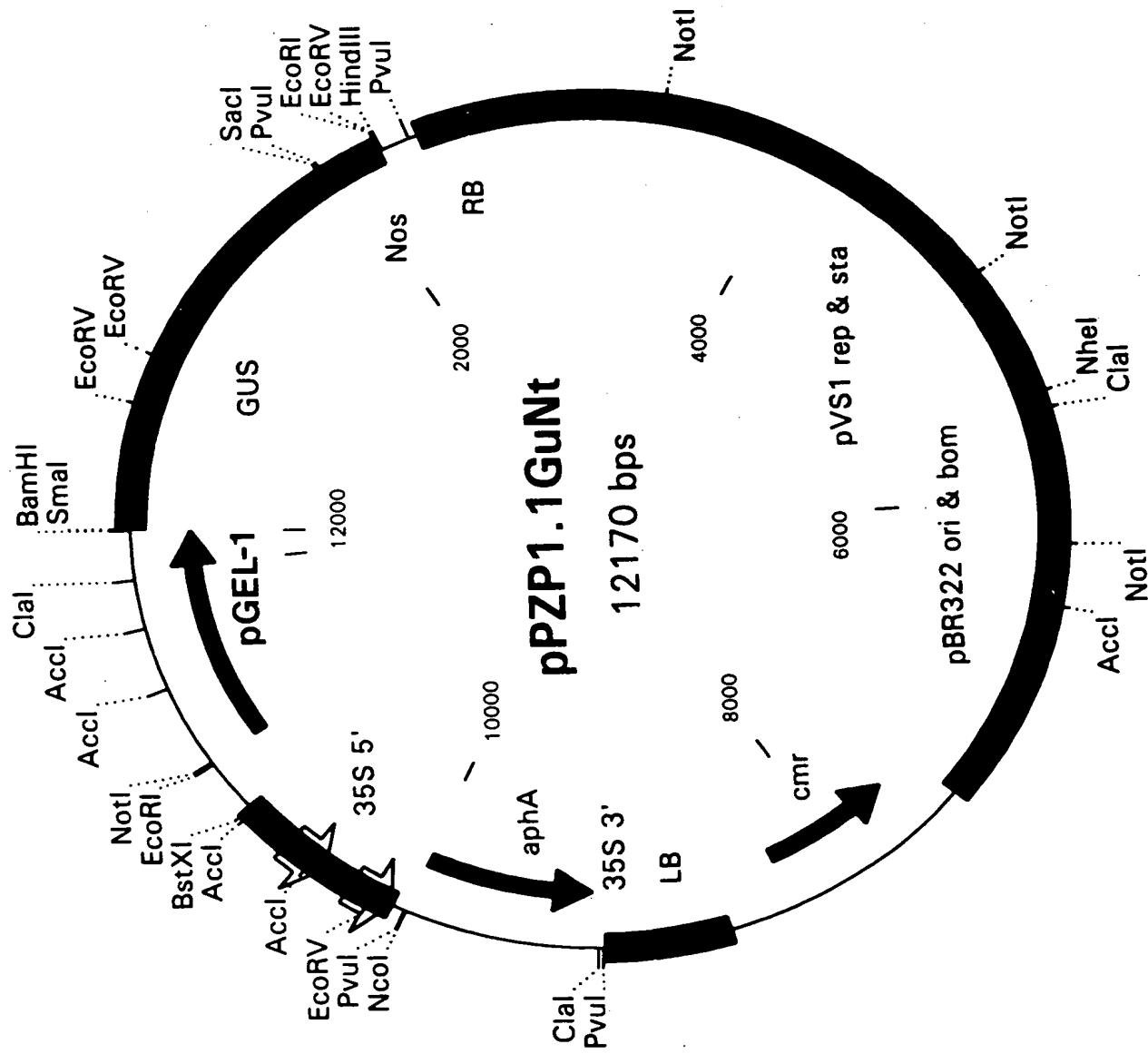
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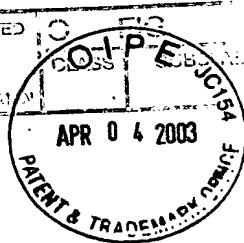
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TECH CENTER 1600/2000

19/28

FIGURE 6A(xii)





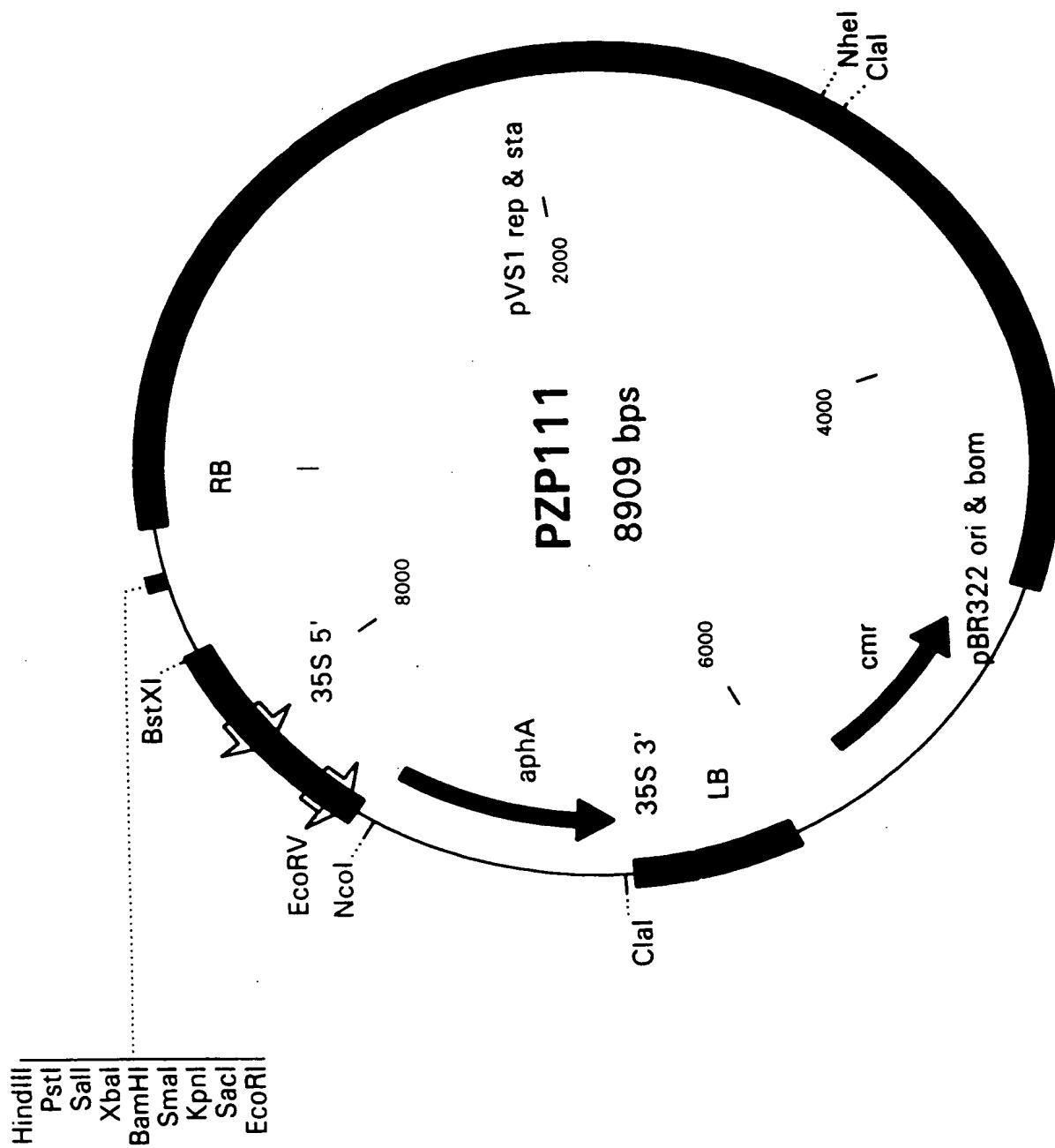
20/28

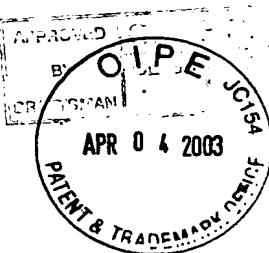
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FIGURE 6B





21 / 28

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APR 09 2003

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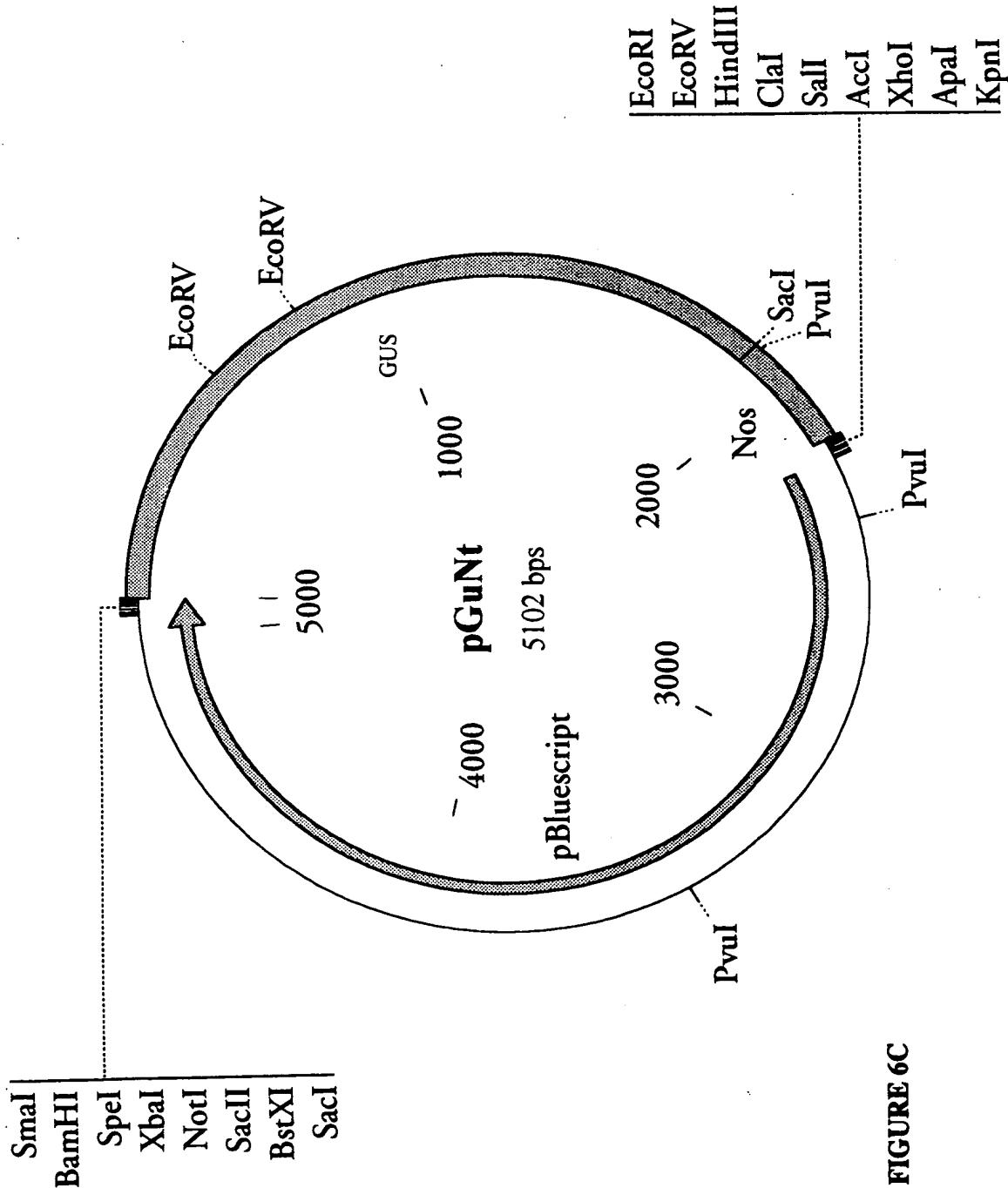
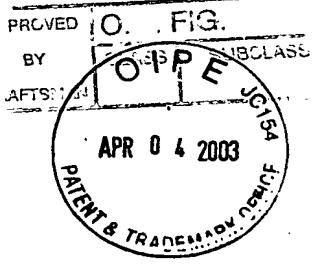


FIGURE 6C



Serial No.: 09/763,957
Docket No.: 229752001300
Inventor: Rose Ramon Botella MESA et al.
Title: A PLANT PROMOTER AND USES THEREFOR
(as amended)
Substitute Sheet 22 of 28

22 / 28

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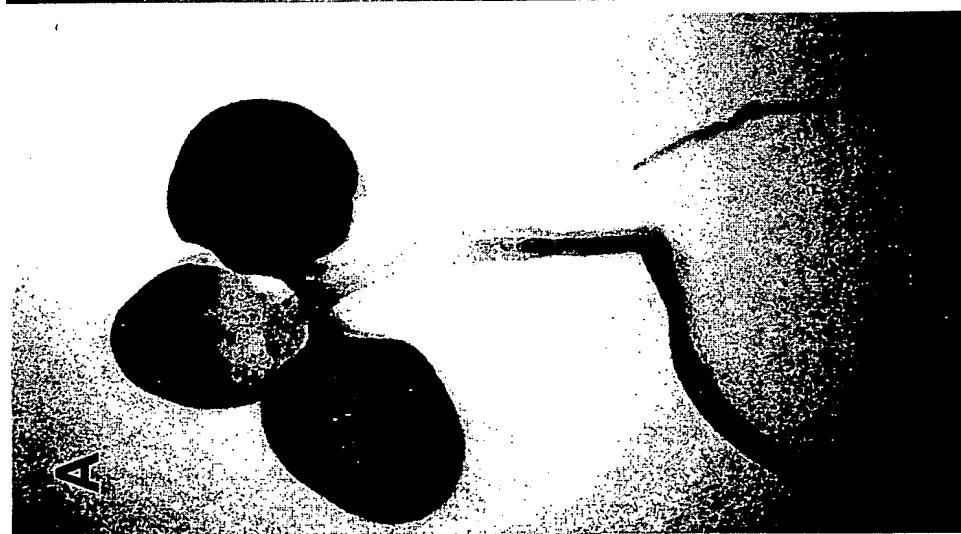
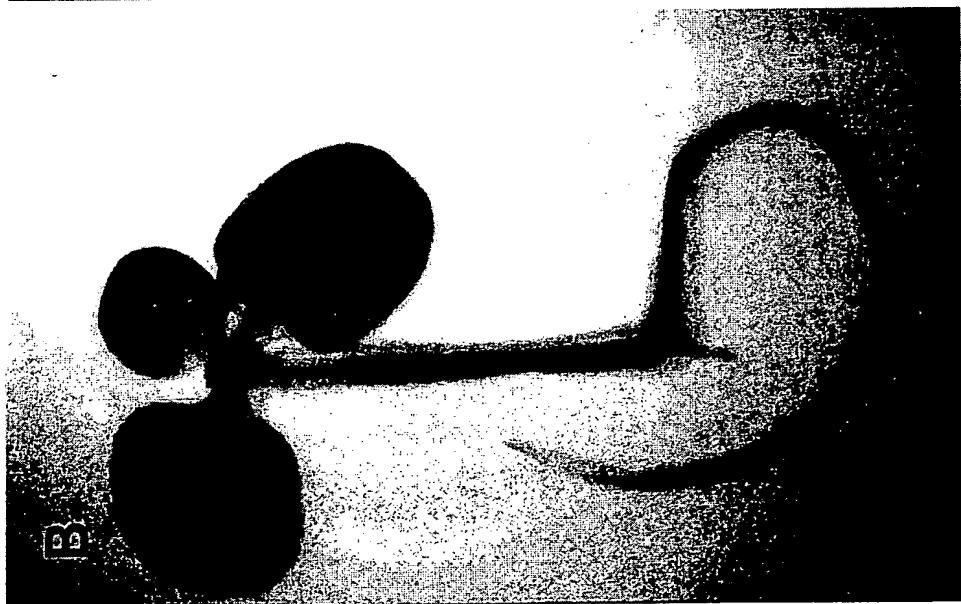
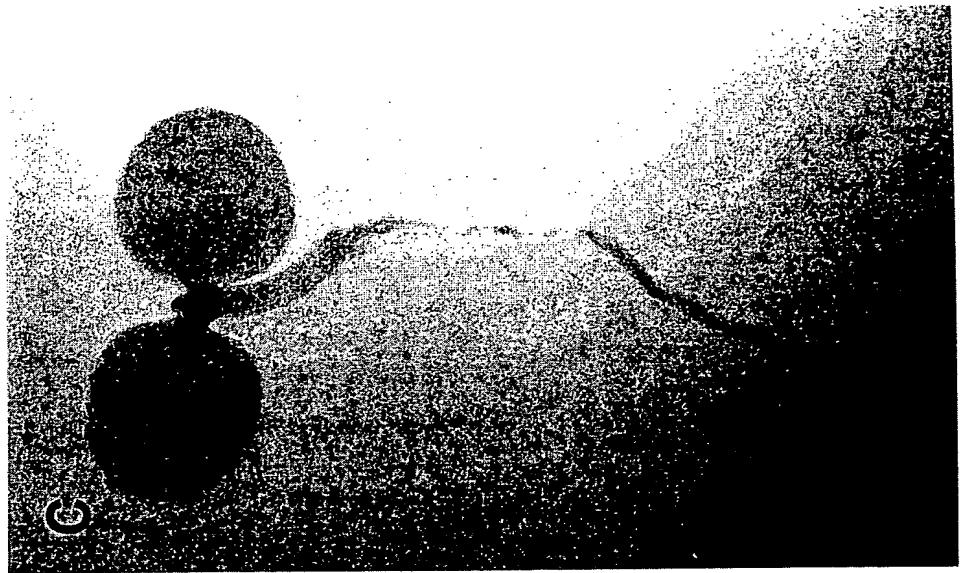


FIGURE 7

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APR 09 2003
TECH CENTER 1633
C-34

23/28

FIGURE 8A

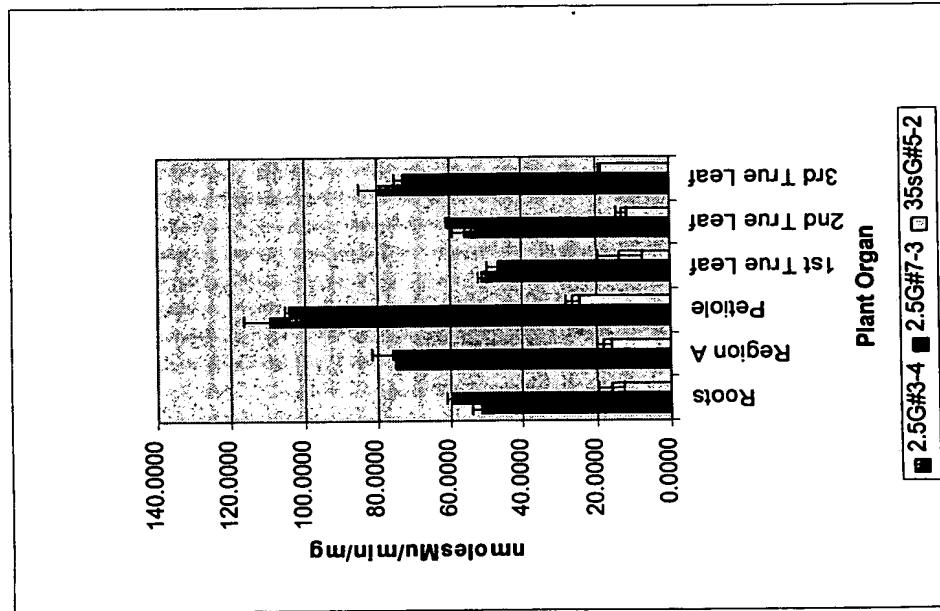
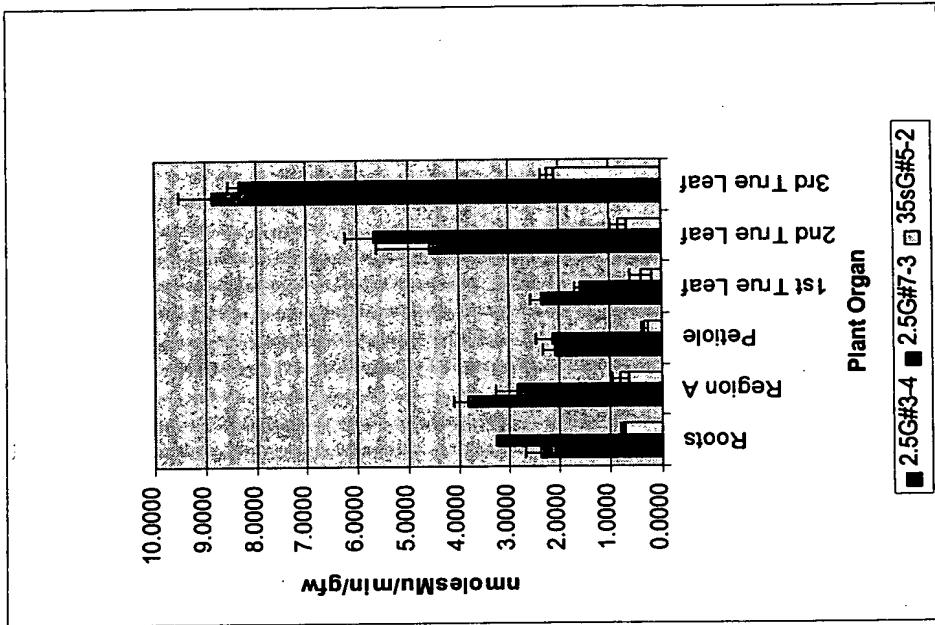


FIGURE 8B





24/28

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 APR 09 2003
 TECH CENTER 1600
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FIGURE 9A

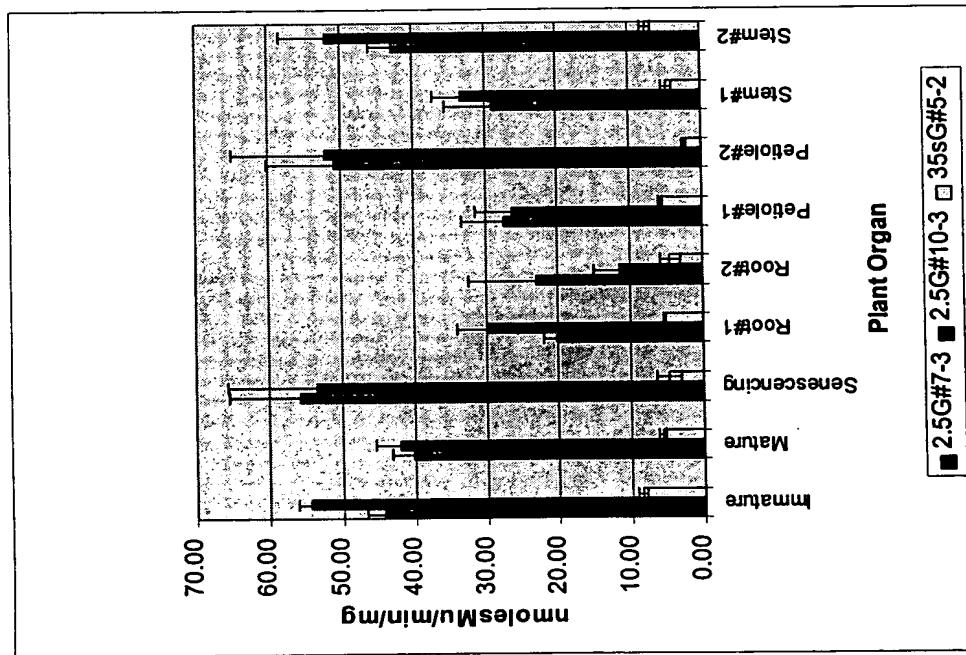
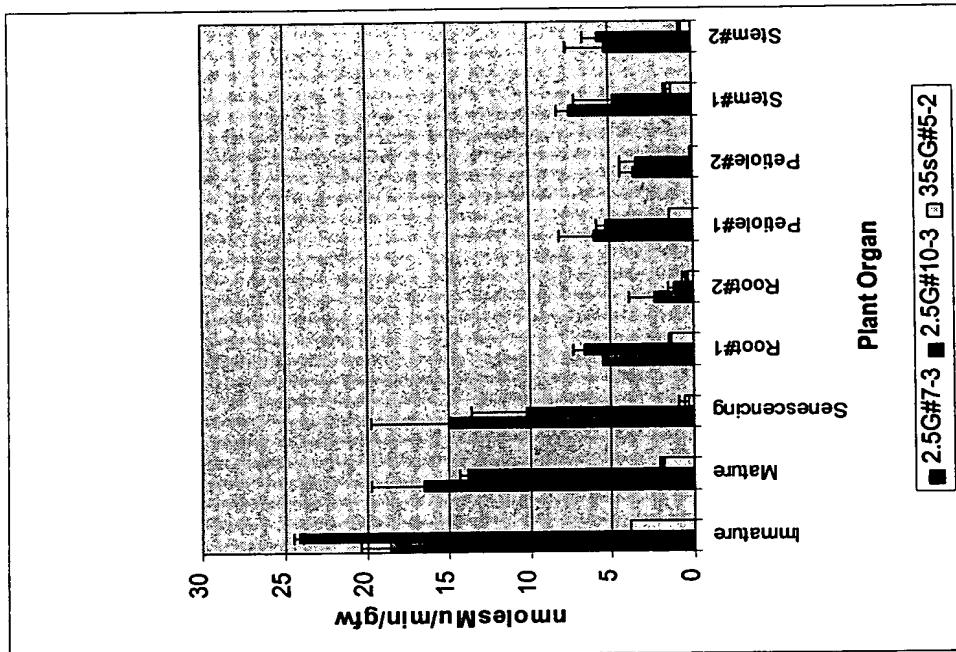


FIGURE 9B





25 / 28

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FIGURE 10A

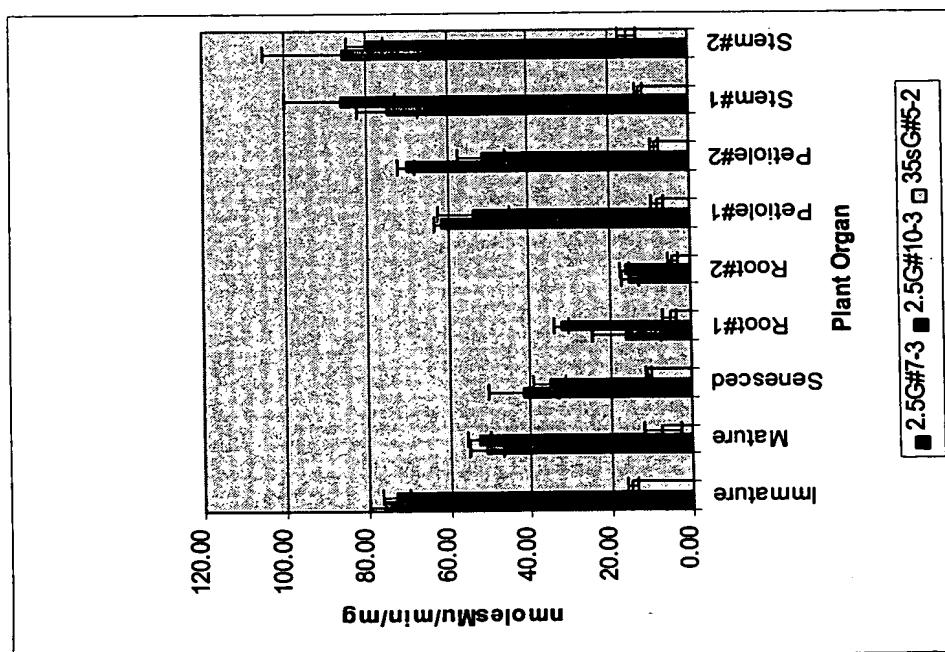
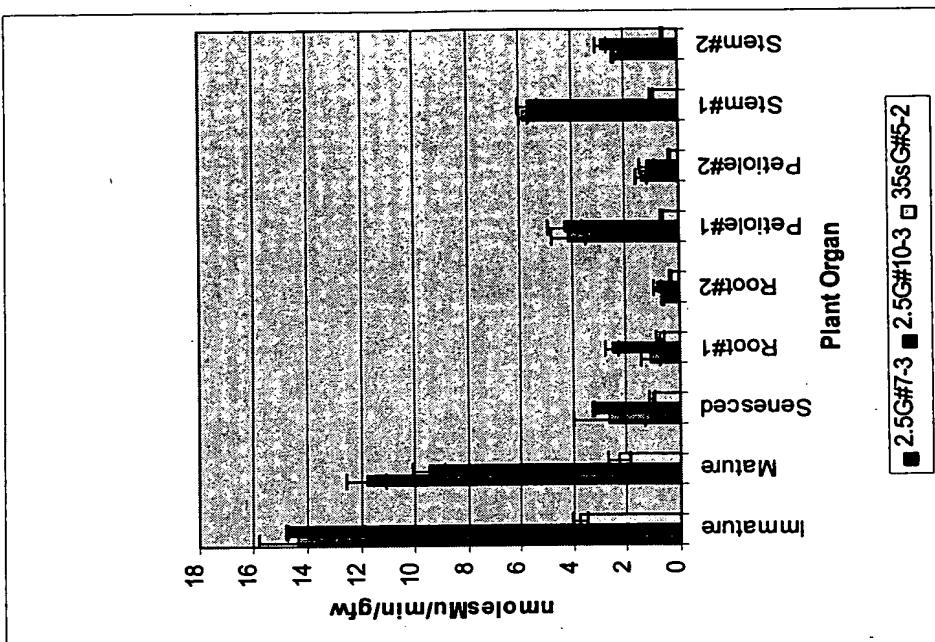
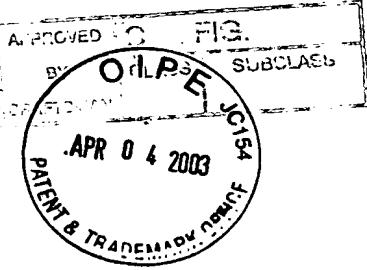


FIGURE 10B





Serial No.: 09/763,957
Docket No.: 229752001300
Inventor: Jose Ramon Botella MESA et al.
Title: A PLANT PROMOTER AND USES THEREFOR
(as amended)
Substitute Sheet 26 of 28

26/28

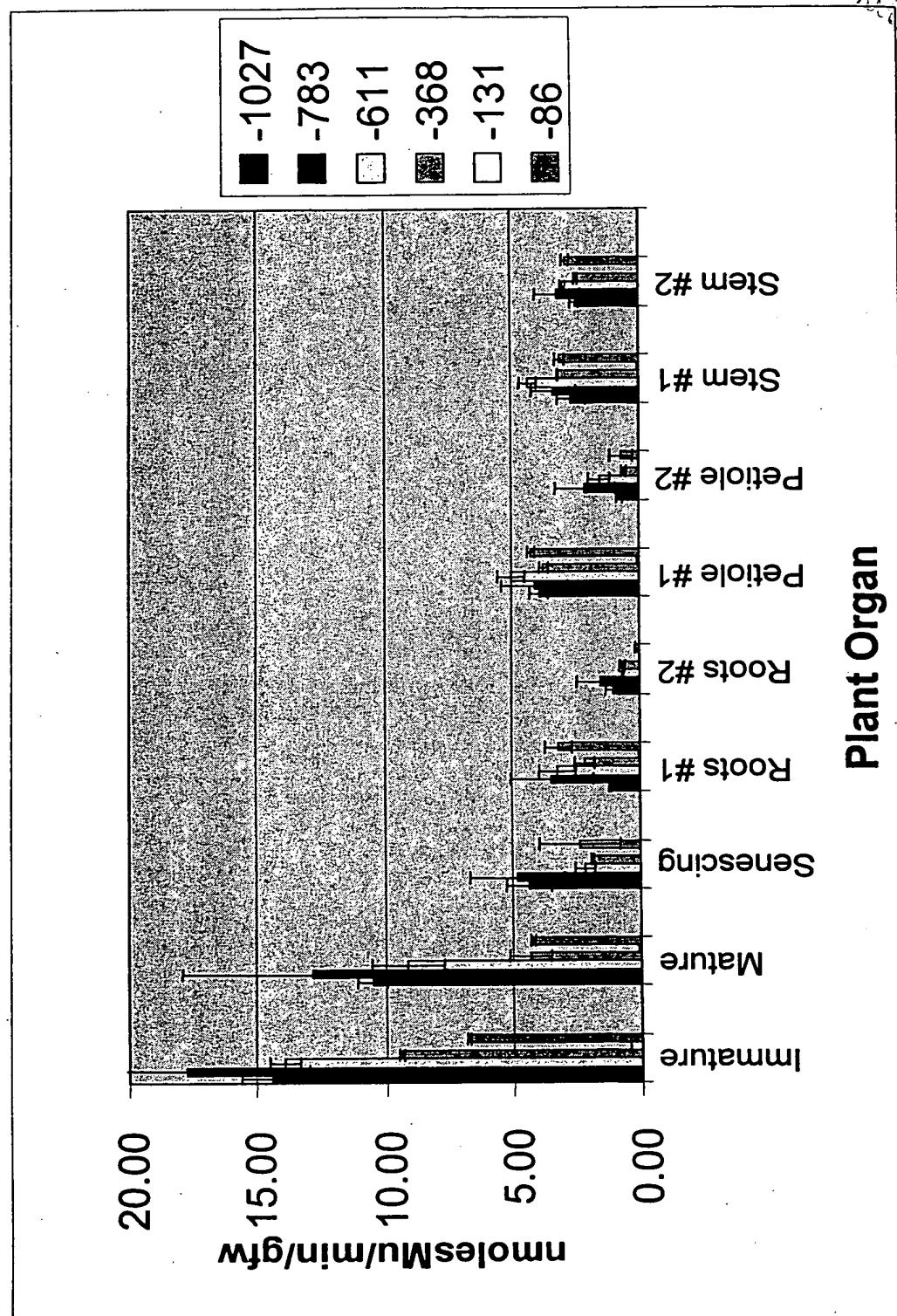
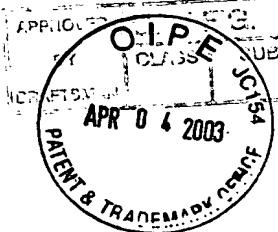
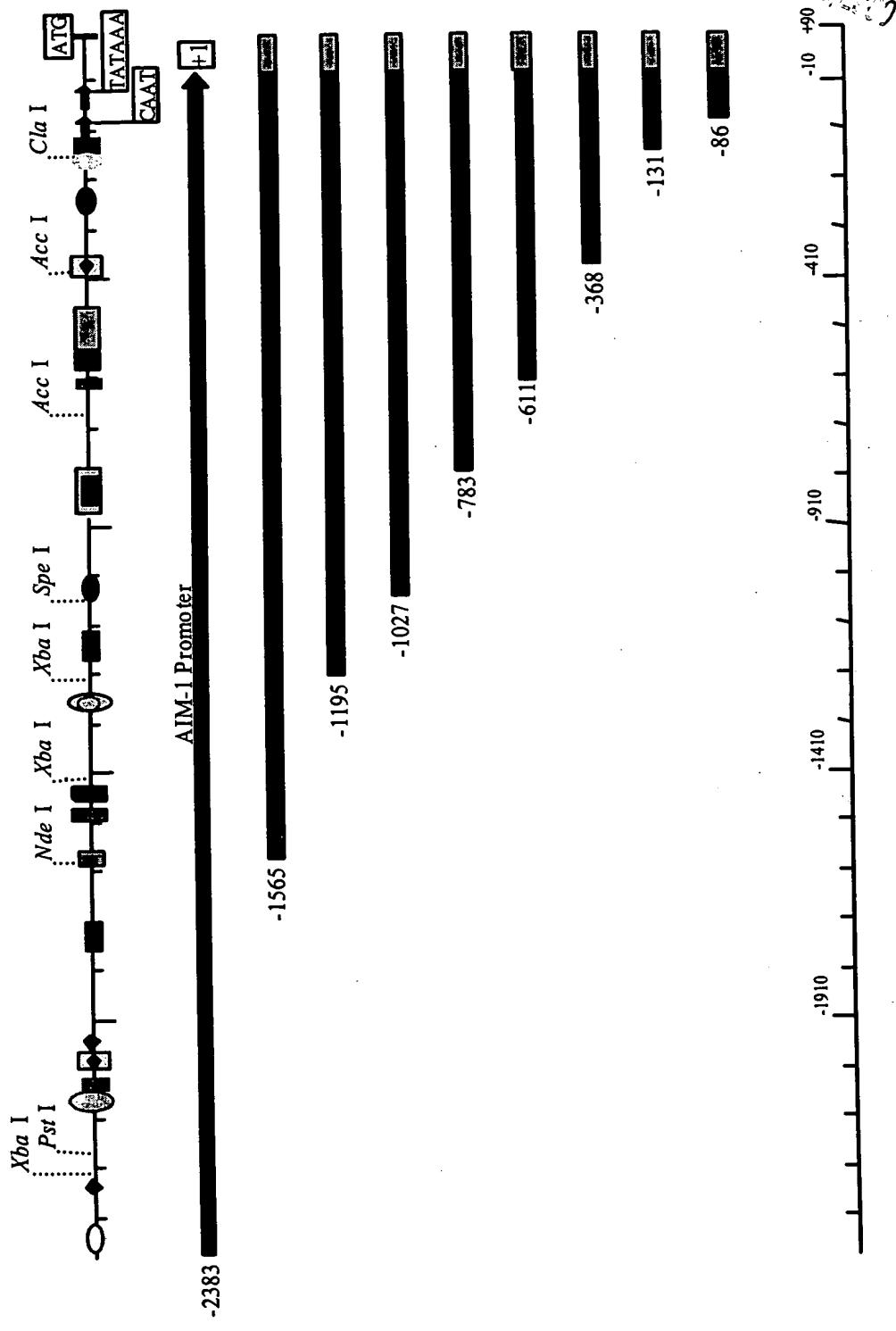


FIGURE 11

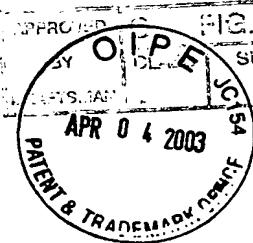


27 / 28

FIGURE 12



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APR 09 2003
TECH CENTER 1632/9379



28/28

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APR 09 2003

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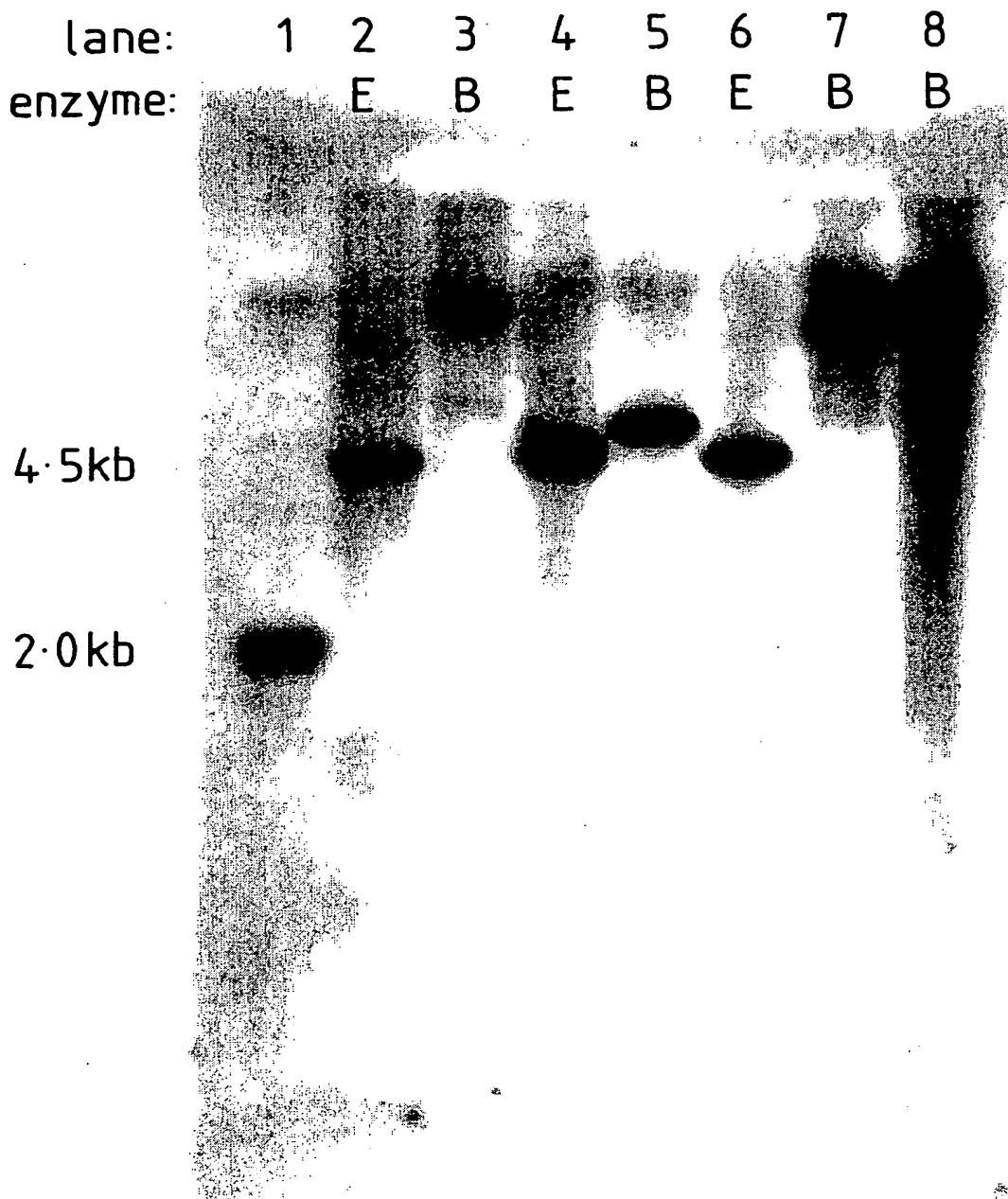


FIGURE 13